

What is claimed is:

1. A method of cell culture comprising:
 - confining a cell between first and second barriers, said barriers spaced at a distance comparable to the size of said cell to contact said cell and prevent said cell from traveling toward or away from each of said first and second barriers; and
 - providing to said cell a culture substance.
2. The method of claim 1 further comprising placing one or more spacers between said first and second barriers to prevent said first and second barriers over-compressing said cell.
3. The method of claim 2 wherein said one or more spacers are rigid spheres, each having a diameter comparable to the size of said cell.
4. The method of any one of claims 1 to 3 wherein each of said barriers has one or more characteristics selected to mimic the characteristics of the biological niche of said cell.
5. The method of any one of claims 1 to 4, wherein said providing comprises establishing a pre-selected concentration gradient of said culture substance within said space.
6. The method of any one of claims 1 to 5, further comprising manually positioning a specific cell type adjacent said cell.
7. The method of any one of claims 1 to 6, further comprising monitoring said cell.
8. The method of any one of claims 1 to 7 wherein a monolayer of cells are cultured between said barriers.
9. The method of claim 8, further comprising removing a cell from said monolayer of cells when said cell to be removed is observed to meet one or more pre-determined criteria.

10. The method of claim 9, wherein said one or more criteria are related to one or more of karyotype, morphology, and size.

11. A method for controlling properties of individual cells in a cell culture, comprising:

confining a cell in a space defined by at least two surfaces that contact said cell and having a size comparable to the size of said cell, each of said at least two surfaces which said cell contacts having one or more pre-selected characteristics;

limiting the number of cells surrounding said cell to permit control of properties of said cell; and

providing a culture substance to said space.

12. The method of claim 11 wherein said pre-selected characteristics are selected to mimic the characteristics of the biological niche of said cell.

13. The method of claim 11 or claim 12 further comprising manually positioning a specific cell type adjacent said cell.

14. The method of any one of claims 11 to 13 further comprising monitoring said cell.

15. The method of any one of claims 11 to 14 wherein a plurality of cells including said cell is cultured as a monolayer.

16. The method of any one of claims 11 to 15 wherein said space has a shape mimicking the shape of the biological niche of said cell.

17. The method of any one of claims 11 to 16, wherein said providing comprises flowing said culture substance to said space at a predetermined rate.

18. The method of claim 17, wherein said culture substance has a pre-selected concentration gradient within said space.

19. The method of any one of claims 11 to 18, wherein said providing comprises allowing said culture substance to permeate through one or more of said at least two surfaces into said space.
20. The method of claim 14, wherein said monitoring comprises sensing, with a sensor disposed adjacent said space, one or more of molecular concentration, temperature, osmolarity, pH, and shear force.
21. The method of any one of claims 11 to 20, wherein said at least two surfaces comprise different types of molecules.
22. A method of forming an artificial tissue, comprising laying a first monolayer of cells on a second monolayer of cells and permitting cells of said first monolayer to interact with cells of said second monolayer wherein each one of said first and second monolayers is cultured according to the method of any one of claims 1 to 21.
23. A combination of a cell culture device and a cell culture, comprising:
 - a container defining a chamber for receiving a fluid culture medium;
 - at least two barriers defining a space in said chamber;
 - a cell constrained in said space and in continuous contact with each one of said barriers during culturing wherein each of said barriers has one or more pre-selected characteristics; and
 - means for providing to said space a culture substance at a predetermined rate.
24. The combination of claim 23 wherein said means for providing to said space a culture substance comprises one or more fluid passageways allowing fluid communication to and from said space.
25. The combination of claim 24, wherein said one or more fluid passageways comprise one or more microfluidic channels each terminating adjacent said space.
26. The combination of any one of claims 23 to 25 further comprising means for regulating fluid flow to or from said space.

27. The combination of any one of claims 23 to 26, wherein at least one of said at least two barriers is permeable to nutrients and gases.
28. The combination of any one of claims 23 to 27 further comprising means for monitoring said cell constrained in said space.
29. The combination of claim 28 wherein said means for monitoring comprises a sensor disposed in said chamber.
30. The combination of claim 29 wherein said sensor is a sensor for sensing one or more of molecular concentration, temperature, osmolarity, pH, and shear force.
31. The combination of claim 29 or claim 30, further comprising one or more transparent electrodes for connecting said sensor to a control system.
32. The combination of any one of claims 23 to 31, wherein at least a portion of one of said barriers is transparent.
33. The combination of claim 32, wherein said barriers comprise a microscope cover slip.
34. The combination of claim 32, wherein said portion of said barrier is made of one of polystyrene, porous glass, or other contact lens materials.
35. The combination of any one of claims 23 to 34, wherein at least one of said barriers is moveable for adjusting the size of said space.
36. The combination of claim 35, further comprising an actuator for moving said at least one of said barriers.
37. The combination of claim 36, wherein said actuator comprises one or more of an inflatable bladder, a screw, a lever, a clamp, a micrometer, and a piezoelectric crystal.

38. The combination of any one of claims 23 to 37, further comprising one or more spacers placed between said barriers for preventing said barriers over-compressing said cell.
39. The combination of claim 38, wherein said one or more spacers are molded on one or more of said barriers.
40. The combination of claims 23 to 39, further comprising a divider for dividing said chamber into a plurality of regions and for preventing said cell from moving between said regions.
41. The combination of claim 40, wherein said divider is removable from said container.
42. The combination of any one of claims 23 to 41, wherein the surfaces of said barriers comprise different types of molecules.
43. The combination of any one of claims 23 to 42, comprising a permeable membrane positioned to cover an opening adjacent said space for preventing said cell from leaving said space through said opening.
44. The combination of any one of claims 23 to 43, wherein said barriers defining a plurality of spaces for confining a plurality of cells therebetween.
45. The combination of any one of claims 23 to 44, which is included in a cartridge.
46. The combination of any one of claims 23 to 45, further comprising said fluid culture medium which is received in said chamber and immerses said cell.
47. The combination of claim 46, wherein at least one wall of said container has a septum allowing access to said space with a syringe or a pipette.
48. The combination of any one of claims 23 to 47, further comprising a capillary conduit for transporting a cell-containing fluid to or from said space.

49. A combination of a cell culturing device and a cell culture, comprising:
- a container defining a chamber for receiving a fluid culture medium;
 - at least two barriers defining a space in said chamber, each one of said barriers having one or more pre-selected characteristics;
 - an assembly of two or more cells constrained in said space so as to keep said assembly therein and in continuous contact with each said barriers during culturing; and
 - means for providing to said space a culture substance at a predetermined rate
50. The combination of claim 49 wherein said space is sufficiently small to permit control of properties of individual cells in culture.
51. The combination of claim 49 or claim 50 wherein said assembly comprises a monolayer of cells.
52. The combination of any one of claims 49 to 51 wherein said means for providing to said space a culture substance comprises one or more fluid passageways allowing fluid communication to and from said space.
53. The combination of claim 52, wherein said one or more fluid passageways comprise one or more microfluidic channels terminating adjacent said space.
54. The combination of any one of claims 49 to 53 further comprising means for regulating fluid flow to or from said space.
55. The combination of any one of claims 49 to 54, wherein at least one of said at least two barriers is permeable to nutrients and gases.
56. The combination of any one of claims 49 to 55 further comprising means for monitoring said cell constrained in said space.
57. The combination of claim 56 wherein said means for monitoring comprises a sensor disposed in said chamber.

58. The combination of claim 57 wherein said sensor is a sensor for sensing one or more of molecular concentration, temperature, osmolarity, pH, and shear force.

59. The combination of claim 57 or claim 58, further comprising one or more transparent electrodes for connecting said sensor to a control system.

60. The combination of any one of claims 49 to 59, wherein at least a portion of one of said barriers is transparent.

61. The combination of claim 60, wherein one of said barriers is a microscope cover slip.

62. The combination of claim 61, wherein said portion of said barrier is made of one of polystyrene, porous glass, or other contact lens materials.

63. The combination of any one of claims 49 to 62, wherein at least one of said barriers is moveable for adjusting the size of said space.

64. The combination of claim 63, further comprising an actuator for moving said at least one of said barriers.

65. The combination of claim 64, wherein said actuator comprises one or more of an inflatable bladder, a screw, a lever, a clamp, a micrometer, and a piezoelectric crystal.

66. The combination of any one of claims 49 to 65, further comprising one or more spacers placed between said barriers for preventing said barriers over-compressing said cell.

67. The combination of claim 66, wherein said one or more spacers are molded on one or more of said barriers.

68. The combination of claims 49 to 67, further comprising a divider for dividing said chamber into a plurality of regions and for preventing said cell from moving between said regions.

69. The combination of claim 68, wherein said divider is removable from said container.
70. The combination of any one of claims 49 to 69, wherein the surfaces of said barriers comprise different types of molecules.
71. The combination of any one of claims 49 to 70, comprising a permeable membrane positioned to cover an opening adjacent said space for preventing a cell from leaving said space through said opening.
72. The combination of any one of claims 49 to 71, wherein said barriers defining a plurality of spaces for confining a plurality of assembly of cells therebetween.
73. The combination of any one of claims 49 to 72, which is included in a cartridge.
74. The combination of any one of claims 49 to 73, further comprising said fluid culture medium which is contained in said chamber and immerses said assembly of cells.
75. The combination of claim 74, wherein at least one wall of said container has a septum allowing access to said space with a syringe or a pipette.
76. The combination of any one of claims 49 to 75, further comprising a capillary conduit for transporting a fluid to or from said space.
77. An assembly of cells comprising a cell cultured in accordance with the method of any one of claims 1 to 21.
78. A cell cultured in accordance with the method of any one of claims 1 to 21.
79. The assembly of cells of claim 77 or the cell of claim 78 for use as an artificial tissue, organ, cell transplant, or *in vitro* fertilization.
80. An apparatus for culturing cells in a controlled artificial niche, comprising:
a container defining a chamber for receiving a fluid culture medium;

at least two barriers defining a space in said chamber for cell culture, the inner surface of each one of said barriers having one or more pre-selected characteristics, wherein said space is sufficiently small to permit control of properties of one or more individual cells cultured in said space; and means for providing a culture substance to said space.

81. The apparatus of claim 80 wherein said barriers comprise two opposing glass plates.

82. The apparatus of claim 80 or claim 81, wherein said space has a size comparable to the size of a single cell.

83. The apparatus of any one of claims 80 to 82, wherein said space limits cells cultured therein to a monolayer.

84. The apparatus of any one of claims 80 to 83 wherein said means for providing comprises one or more fluid passageways allowing fluid communication to and from said space.

85. The apparatus of claim 84 wherein said one or more fluid passageways comprise one or more microfluidic channels terminating adjacent said space.

86. The apparatus of any one of claims 80 to 85 further comprising means for regulating fluid flow to or from said space.

87. The apparatus of any one of claims 80 to 86 wherein at least one of said barriers is permeable to nutrients and gases.

88. The apparatus of any one of claims 80 to 87 further comprising means for monitoring a cell constrained in said space.

89. The apparatus of claim 88 wherein said means for monitoring comprises a sensor disposed in said chamber.

90. The apparatus of claim 89 wherein said sensor is a sensor for sensing one or more of molecular concentration, temperature, osmolarity, pH, and shear force.

91. The apparatus of claim 89 or claim 90, further comprising one or more transparent electrodes for connecting said sensor to a control system.
92. The apparatus of any one of claims 80 to 91, wherein at least a portion of one of said barriers is transparent.
93. The apparatus of claim 92, wherein one of said barriers is a microscope cover slip.
94. The apparatus of claim 93, wherein said portion of said barrier is made of one of polystyrene, porous glass, or other contact lens materials.
95. The apparatus of any one of claims 80 to 94, wherein at least one of said barriers is moveable for adjusting the size of said space.
96. The apparatus of claim 95, further comprising an actuator for moving said at least one of said barriers.
97. The apparatus of claim 96, wherein said actuator comprises one or more of an inflatable bladder, a screw, a lever, a clamp, a micrometer, and a piezoelectric crystal.
98. The apparatus of any one of claims 80 to 97, further comprising one or more spacers placed between said barriers for preventing said barriers over-compressing a cell cultured therebetween.
99. The apparatus of claim 98, wherein said one or more spacers are molded on one or more of said barriers.
100. The apparatus of claims 80 to 99, further comprising a divider for dividing said chamber into a plurality of regions and for preventing said cell from moving between said regions.
101. The apparatus of claim 100, wherein said divider is removable from said container.

102. The apparatus of any one of claims 80 to 101, wherein the surfaces of said barriers comprise different types of molecules.

103. The apparatus of any one of claims 80 to 102, comprising a permeable membrane positioned to cover an opening adjacent said space for preventing a cell from leaving said space through said opening.

104. The apparatus of any one of claims 80 to 103, wherein said barriers defining a plurality of spaces for confining a plurality of cells therebetween.

105. The apparatus of any one of claims 80 to 104, which is included in a cartridge.

106. The apparatus of any one of claims 80 to 105, further comprising said fluid culture medium which is contained in said chamber.

107. The apparatus of claim 106, wherein at least one wall of said container has a septum allowing access to said space with a syringe or a pipette.

108. The apparatus of any one of claims 80 to 107, further comprising a capillary conduit for transporting a fluid to or from said space.

109. An assembly for cell culturing comprising a plurality of devices, each one of said devices being one of the combination of any one of claims 23 to 79 and the apparatus of any one of claims 80 to 108.

110. The assembly of claim 109, wherein each one of said plurality of devices is generally plate-shaped and said plurality of devices are stacked in parallel.

111. A method of cell culturing, comprising:

culturing one or more cells while restricting movement of said one or more cells such that each one of said one or more cells is in continuous contact with two opposing barrier surfaces and is mobile between said barrier surfaces.

112. The method of claim 111, wherein said barrier surfaces are generally parallel with each other.

113. The method of claim 111 or claim 112, wherein said barrier surfaces are generally planar.

114. The method of any one of claims 111 to 113, further comprising, during said culturing, obtaining one or more images of said one or more cells.

115. The method of claim 114, wherein said one or more images are obtained using a non-confocal imaging device.

116. The method of claim 114, wherein said one or more images are obtained using a bright field imaging device or a fluorescent imaging device.

117. The method of claim 116, wherein said one or more images are obtained using a differential interference contrast (DIC) imaging device.

118. The method of any one of claims 114 to 117, wherein said one or more images comprise a plurality of images taken over a period of time longer than about one day.

119. The method of any one of claims 114 to 117, wherein said one or more images comprise a plurality of images taken over a period of time which is not limited by significant or substantial phototoxic effects to impose a constraint for gathering information on behavior histories of a cell or cells.